

IN THE CLAIMS

1-17. Cancelled

18. (Currently Amended) A method for quantitating newly initiated RNA of a positive strand RNA virus comprising:

contacting an isolated replicase complex for the positive strand RNA virus, an isolated viral replicon template RNA for the positive strand RNA virus, nucleotides, and a labeled nucleotide analog, and 2'-O-methyl-5-methyluridine-5'- triphosphate under conditions sufficient for *in vitro* RNA synthesis, to form a newly synthesized RNA population comprising the labeled nucleotide analog;

hybridizing a probe and the newly synthesized RNA population comprising the labeled nucleotide analog, under stringent hybridization conditions, wherein the probe is complementary to at least a portion of a transcription initiation region of the newly synthesized RNA population;

digesting unhybridized, single-stranded RNA with a single-strand specific ribonuclease to form a protected RNA population comprising the labeled nucleotide analog;

detecting the protected RNA population comprising the labeled nucleotide analog; and

quantitating the protected RNA population comprising the labeled nucleotide analog.

19. Cancelled

20. (Previously Presented) The method of Claim 18, further comprising providing the isolated replicase complex and the isolated viral replicon template RNA by:

transfecting a human hepatoma cell line with a viral replicon RNA or a DNA template for a viral replicon to provide a transfected cell line,

incubating the transfected cell line under conditions suitable for production of viral replicase complexes, and

isolating the replicase complexes comprising the viral replicon template RNA from the cell membrane fraction of the transfected cells.

21. (Original) The method of Claim 20, wherein the DNA template for the viral replicon is SEQ ID NO: 1, SEQ ID NO: 2, SEQ ID NO:3, SEQ ID NO: 4, or SEQ ID NO: 5.

22. Cancelled

23. (Original) The method of Claim 18, wherein the labeled nucleotide analog is an analog capable of being recognized by a specific antibody, an analog which can be recognized via a high specificity binding reaction, or an analog directly detectable as a result of a physical property of the analog.

24 - 30. Cancelled

31. (Previously Presented) A method for determining whether a test compound is an RNA synthesis initiation inhibitor of a positive strand RNA virus comprising:

contacting an isolated replicase complex for the positive strand RNA virus, an isolated viral replicon template RNA for the positive strand RNA virus, nucleotides, a labeled nucleotide analog, and the test compound, under conditions sufficient for *in vitro* RNA synthesis, to form a newly synthesized RNA population comprising the labeled nucleotide analog;

hybridizing a probe and the newly synthesized RNA population comprising the labeled nucleotide analog, under stringent hybridization conditions, wherein the probe is complementary to at least a portion of an initiation region of the newly synthesized RNA population;

digesting unhybridized, single-stranded RNA with a single-strand specific ribonuclease to form a protected RNA population comprising the labeled nucleotide analog;

detecting the protected RNA population comprising the labeled nucleotide analog;

quantitating the protected RNA population comprising the labeled nucleotide analog to provide a test RNA amount; and

comparing the test RNA amount with a control RNA amount of protected RNA comprising the labeled nucleotide analog but produced in the absence of the test compound, wherein a decrease in the test RNA amount compared to the control RNA amount indicates that the test compound inhibits RNA synthesis initiation of the positive strand RNA virus.

32. Cancelled

33. (Previously Presented) The method of Claim 31, further comprising providing the isolated replicase complex and the isolated viral replicon template RNA by:
transfecting a human hepatoma cell line with a viral replicon RNA or a DNA template for a viral replicon to provide a transfected cell line,
incubating the transfected cell line under conditions suitable for production of viral replicase complexes, and
isolating the replicase complexes comprising the viral replicon template RNA from the cell membrane fraction of the transfected cells.

34. – 35. Cancelled

36. (Original) The method of Claim 31, wherein the positive strand RNA virus is Hepatitis C Virus.

37. – 50. Cancelled

51. (Previously Presented) The method of Claim 18, wherein the positive strand RNA virus is Hepatitis C Virus.

52. (New) A method for determining whether a test compound is an RNA synthesis initiation inhibitor of a positive strand RNA virus comprising:
contacting an isolated replicase complex for the positive strand RNA virus, an isolated viral replicon template RNA for the positive strand RNA virus, nucleotides, a labeled nucleotide analog, 2'-O-methyl-5-methyluridine-5'- triphosphate, and the test compound, under conditions sufficient for *in vitro* RNA synthesis, to form a newly synthesized RNA population comprising the labeled nucleotide analog;
hybridizing a probe and the newly synthesized RNA population comprising the labeled nucleotide analog, under stringent hybridization conditions, wherein the probe is complementary to at least a portion of an initiation region of the newly synthesized RNA population;
digesting unhybridized, single-stranded RNA with a single-strand specific ribonuclease to form a

protected RNA population comprising the labeled nucleotide analog;

detecting the protected RNA population comprising the labeled nucleotide analog;

quantitating the protected RNA population comprising the labeled nucleotide analog to provide a test RNA amount; and

comparing the test RNA amount with a control RNA amount of protected RNA comprising the labeled nucleotide analog but produced in the absence of the test compound, wherein a decrease in the test RNA amount compared to the control RNA amount indicates that the test compound inhibits RNA synthesis initiation of the positive strand RNA virus.